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EXPERIMENTAL STUDIES

Long-Term Beta-Blocker Treatment Prevents Chronic Creatine Kinase and Lactate Dehydrogenase System Changes in Rat Hearts After Myocardial Infarction

ANNE LASER, PHD, STEFAN NEUBAUER, MD,* RONG TIAN, MD,† KAI HU, MD, PETER GAUDRON, MD, JOANNE S. INGWALL, PHD,† GEORG ERTL, MD

Mannheim and Würzburg, Germany and Boston, Massachusetts

Objectives. **We tested the hypothesis that long-term beta-blocker treatment with bisoprolol prevents creatine kinase (CK) and lactate dehydrogenase system changes that occur after chronic myocardial infarction.**

Background. **The mechanism of the beneficial effect of beta-blocker therapy is still unclear.**

Methods. **Six groups of rats were studied. Sham operated (sham) and hearts with ligated left anterior descending coronary artery (myocardial infarction) were untreated, treated early (beginning 30 min after infarction) or treated late (beginning 14 days after infarction). After 8 weeks, hearts were isolated and buffer perfused isovolumetrically. With a left ventricular balloon, mechanical function was recorded at an end-diastolic pressure of 10 mm Hg. Biopsy samples of noninfarcted left ventricular tissue were taken. Enzyme activities were measured spectrophotometrically; isoenzymes were separated by agar gel electrophoresis; and total creatine levels were measured with high performance liquid chromatography.**

Results. **The decrease in left ventricular developed pressure in untreated hearts (120 ± 9 vs. 104 ± 5 mm Hg [mean ± SE], $p < 0.05$, sham vs. myocardial infarction) after myocardial infarction was prevented by early treatment (118 ± 9 vs. 113 ± 4 mm Hg). Late treatment failed to improve mechanical function. Reduction of CK activity occurring in untreated infarcted hearts (6.4 ± 0.3 vs. 5.1 ± 0.3 IU/mg protein, $p < 0.05$, sham vs. myocardial infarction) was prevented by early beta-blocker therapy. The increase in CK isoenzyme BB and MB levels, decrease in mitochondrial CK isoenzyme levels and increase in anaerobic lactate dehydrogenase isoenzyme levels in untreated infarcted hearts did not occur during bisoprolol treatment. The decrease in total creatine levels after myocardial infarction (74.2 ± 4.9 vs. 54.9 ± 3.3 nmol/mg protein, $p < 0.05$, sham vs. myocardial infarction) was prevented by bisoprolol treatment. Early treatment was more effective than late therapy in preventing CK and lactate dehydrogenase system changes. In addition, in sham hearts, a 40% increase of creatine levels above normal levels was detected.**

Conclusions. **Bisoprolol prevented changes in CK and lactate dehydrogenase systems that occur after myocardial infarction. These observations may be related to the beneficial effects of long-term beta-blocker treatment in patients with chronic myocardial infarction.**

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In recent large clinical trials, mortality was reduced by beta-adrenergic blocking agents in patients with heart failure resulting from dilated cardiomyopathy but not in patients with a history of myocardial infarction (1). In contrast, in patients treated for myocardial infarction with beta-blockers, benefit was related to cardiac dysfunction (2). This apparent paradox remains unresolved and underlines our lack of understanding of the mechanisms of beta-blocker therapy in patients with chronic heart failure. Lowering heart rate may be one major beneficial effect (3–5); others may be antiarrhythmic actions (6,7) or up-regulation of beta-receptor density (3,4). Various studies on patients with heart failure showed a hemodynamic benefit of chronic beta-blocker treatment (8). Beta-blockade in heart failure improved exercise capacity, left ventricular ejection fraction, stroke and cardiac index and arteriovenous oxygen difference. Myocardial oxygen consumption was unchanged despite the increase in cardiac mechanical work, implying that beta-blocker therapy was energetically favorable for the heart. However, data are inconclusive, and studies have been hampered by inhomogeneity of diseases underlying heart failure and variation of therapeutic regimens.

The dilated heart faces chronically increased wall stress (9) and energy demand. In an animal model of chronic myocardial infarction, we have previously shown that energy reserve of residual intact myocardium is impaired with substantial reductions of creatine phosphate, total creatine, total creatine kinase (CK) and mitochondrial CK isoenzyme (mito-CK) activity and a shift of lactate dehydrogenase isoenzymes towards anaerobic

From the Würzburg University, Würzburg and *Mannheim/Heidelberg University, Mannheim, Germany; and †Brigham and Women's Hospital, Boston, Massachusetts. This study was supported by Grants SFB 355/A-3 and B-1 from the Deutsche Forschungsgemeinschaft, Bonn, Germany and by a grant from Merck AG, Darmstadt, Germany. Helena Laboratories, Hartheim, Germany supplied the electrophoresis equipment.

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Address for correspondence: Professor Dr. Georg Ertl, II. Medizinische Klinik, Klinikum Mannheim der Universität Heidelberg, 68135 Mannheim, Germany.

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metabolism (10). With the same infarcted rat heart model, the purpose of the present study was to test whether beta-blocker treatment with bisoprolol prevents CK and lactate dehydrogenase system changes. In addition, we tested late treatment versus early treatment after myocardial infarction.

Methods

Animals and experimental myocardial infarction. Left anterior descending coronary artery ligations were induced in 12-week old male Wistar rats as previously described (11). A left thoracotomy was performed under ether anesthesia and positive pressure ventilation. The heart was rapidly exteriorized by applying gentle pressure on both sides of the thorax. The left anterior descending coronary artery was ligated between the pulmonary artery outflow tract and the left atrium. After replacement of the heart into the thorax, the lungs were inflated by increasing positive end-expiratory pressure, and the wound was closed immediately. Sham operations were performed using an identical procedure, except that the suture was passed under the coronary artery without ligation. For the first 24 h, the postoperative mortality rate of infarct group was 40% to 50%. The surviving rats were kept on standard rat chow and water ad libitum. All procedures conformed to the guiding principles of the American Physiological Society.

Study groups. Preliminary experiments were performed to find a hemodynamically effective dose for beta-blocker treatment: 66 mg/kg body weight per day in drinking water decreased heart rate in vivo from 402 ± 24 beats/min (mean ± SE) in untreated rats ($n = 5$) to 321 ± 25 beats/min in bisoprolol-treated rats ($n = 5$, $p < 0.05$). Smaller dosages did not reduce heart rate significantly.

Six groups of rats were studied: untreated rats with sham operation (sham) or left anterior descending coronary artery ligation (myocardial infarction), sham or myocardial infarction rats treated with bisoprolol starting 30 min after infarction (sham B and myocardial infarction B) and sham and myocardial infarction rats in which bisoprolol treatment started 14 days after infarction (sham 14 d B and myocardial infarction 14 d B). During the following 8 weeks, one untreated rat, one rat from the early treatment group and three from the late treatment group died. Thus, 8 weeks postoperatively, we studied 11 untreated sham and 13 untreated myocardial infarction rats, 12 late-treated sham and 13 late-treated myocardial infarction rats and 9 early-treated sham and 14 early-treated myocardial infarction hearts. Bisoprolol treatment was stopped 1 day before isolating the heart to exclude effects of acute beta-blockade on our measurements. Bisoprolol is known to have an elimination half-life of 9 to 12 h (12).

Isolated rat heart preparation. Rats were anesthetized 8 weeks after infarction by injection of 20 mg pentobarbital sodium intraperitoneally. A transverse laparotomy and a left and a right anterolateral thoracotomy were performed. The heart was rapidly excised and immersed in ice-cold buffer. At a constant temperature of 37°C, retrograde perfusion of the heart via the aorta was started in the Langendorff mode at a constant coronary perfusion pressure of 100 mm Hg. Krebs–Henseleit buffer of the following composition was used (in mmol/liter): sodium chloride 118.0, potassium dihydrogen phosphate 1.2, potassium chloride 4.7, magnesium sulfate 1.2, calcium chloride 1.75, disodium EDTA 0.5, glucose 11.0, sodium bicarbonate 25.0. Buffer was freshly made each day and was equilibrated with 95% oxygen + 5% carbon dioxide, yielding a pH of 7.4. The root of the pulmonary artery was cut open to allow for right ventricular outflow. The flow of the Thebesian veins was drained with a small polyethylene tubing pierced through the apex of the left ventricle. The heart was surrounded by its own perfusate in a water-jacketed reservoir. The perfusate level was kept constant immediately above the pulmonary outflow tract by continuous suction.

Cardiac performance measurements. A water-filled latex balloon was inserted into the left ventricle through an incision in the left atrium and was secured by a ligature. The balloon was connected to a Statham P23Db pressure transducer (Gould Instruments) by a small-bore stainless-steel tube. By varying the filling volume of the balloon, an end-diastolic pressure of 10 mm Hg was set; left ventricular pressure and heart rate were continuously recorded on an eight-channel recorder (Polygraph, ZAK).

Determination of infarct size. Because tissue was needed for biochemical measurements, the heart was separated into atria, right ventricle, left ventricular free wall, septum and scar after performance measurements. Each part was weighed. A sample of about 100 mg of intact left ventricular wall was taken for biochemical measurements. Infarcted hearts with a scar weight less than 200 mg were excluded. Each piece of scar was flattened between two glass plates. Photographs were taken with millimeter paper as background to determine scar area. Left ventricular volume was used to calculate left ventricular surface area. Infarct size was calculated as the percent of scar area in surface area. This method was previously shown to correlate closely with infarct size determined by standard morphologic techniques (13).

High performance liquid chromatographic measurements. Tissue was homogenized in 0.42 mol/liter perchloric acid using the Potter S homogenizer (B. Braun Melsungen AG), keeping the sample temperature at 4°C. After aliquots for protein measurements (14) were taken, 1 mol/liter potassium hydroxide was added to set a pH of about 5. The sample was centrifuged, filtered and injected into the high performance liquid chromatographic (HPLC) system for determination of total creatine. The HPLC system consisted of a pump (Waters Model 590, Millipore GmbH), an automatic injection system (Waters 712 WISP, Millipore GmbH), a spectrophotometer (Waters 486 Tunable Absorbance Detector, Millipore GmbH), a recorder (Waters 740 Data Module, Millipore GmbH) and a reversed-phase column (Supelcosil LC-18, 15.0 cm × 4.6 mm, Supelco Deutschland GmbH). The 2-cm guard column had the same stationary phase as the HPLC column: spherical 5-μm particles on a silica core with C-18 coating. The chromatography was run at room temperature and the wavelength of the

Table 1. Infarct Size, Left Ventricular Weight and Body Weight

	Untreated	Bisoprolol Treatment	
		Late	Early
Sham	n = 11	n = 12	n = 9
MI	n = 13	n = 13	n = 14
Infarct size (%)			
Sham	—	—	—
MI	36 ± 2	35 ± 2	36 ± 2
LVW (g)			
Sham	1.06 ± 0.04	0.96 ± 0.04	0.94 ± 0.06
MI	1.10 ± 0.05	0.98 ± 0.03	0.92 ± 0.04*
BW (g)			
Sham	479 ± 25	460 ± 13	457 ± 23
MI	518 ± 14	462 ± 17	446 ± 14*
LVW/BM (mg/g)			
Sham	2.27 ± 0.14	2.09 ± 0.07	2.01 ± 0.07
MI	2.14 ± 0.09	2.13 ± 0.07	2.07 ± 0.08

*$p < 0.05$ treated versus untreated rats. Data presented are mean value ± SE or number of rats. BW = body weight; LVW = left ventricular weight; MI = left anterior descending coronary artery-ligated hearts; Sham = sham-operated hearts.

Table 2. Left Ventricular Developed Pressure and Heart Rate

	Untreated	Bisoprolol Treatment	
		Late	Early
Sham	n = 11	n = 12	n = 9
MI	n = 13	n = 13	n = 14
LVDP (mm Hg)			
Sham	120 ± 9	119 ± 7	118 ± 9
MI	104 ± 5*	105 ± 6*	113 ± 4
HR (beats/min)			
Sham	235 ± 9	240 ± 8	233 ± 11
MI	230 ± 11	222 ± 10	223 ± 6

*$p < 0.05$, left anterior descending coronary artery-ligated hearts (MI) versus sham-operated hearts (Sham). Data presented are mean value ± SE or number of rats. HR = heart rate; LVDP = left ventricular developed pressure at end-diastolic pressure of 10 mm Hg.

detector was set at 206 nm. The mobile phase consisted of potassium dihydrogen phosphate (215 mmol/liter), tetrabutylammoniumhydrogen sulfate (2.3 mmol/liter) and acetonitrile (3.5%). A similar method was described by Sellevold et al. (15).

Enzyme measurements. Each sample (5 to 10 mg of tissue) was homogenized in 0.1 mol/liter phosphate buffer, pH 7.4, containing 1 mmol/liter EGTA and 1 mmol/liter beta-mercaptoethanol. Before 0.1% Triton X was added, aliquots for measurement of protein were taken. All samples were kept on ice. The following enzyme activities were measured using an Ultraspec III spectrophotometer (Pharmacia Biosystems): CK (16), lactate dehydrogenase (17) and citrate synthase (18). The isoenzymes of lactate dehydrogenase were determined with the Titan Gel LD Isoenzyme System (Helena Diagnostika GmbH) using agarose gel electrophoresis. To measure the CK isoenzyme distribution, the Rapid Electrophoresis System (REP, Helena Diagnostika GmbH) was used as separation unit, and the REP CK isoforms kit (Helena Diagnostika GmbH) for agarose gel and incubation solution were used. The agarose gel contains a Tris/barbital buffer with sodium azide as preservative. The Electrophoresis Data Center (EDC, Helena Diagnostika GmbH) automatically quantified the separated isoenzyme bands.

Statistical analysis. All data are presented as mean value ± SE. For comparison of the six groups (sham, myocardial infarction, sham 14d B, myocardial infarction 14d B, sham B and myocardial infarction B), a one-factor analysis of variance (ANOVA) with the Games–Howell test was used as post hoc test. Statistical calculations were guided by the SuperANOVA statistics program (Abacus Concepts, Inc.).

Results

Infarct size, body weight and ventricular weight. Average infarct sizes (Table 1) were comparable for all treatment groups. Infarcted rats in the early-treated group showed reduced body weights (518 ± 14 vs. 446 ± 14 g, $p < 0.05$, myocardial infarction vs. myocardial infarction B) and reduced left ventricular weights (1.10 ± 0.05 vs. 0.92 ± 0.04 g, $p < 0.05$, myocardial infarction vs. myocardial infarction B), but no significant differences occurred in the ratio of left ventricular weight to body weight among groups.

Performance of treated and untreated infarcted hearts. Table 2 shows performance data from treated and untreated infarcted hearts under control conditions at an end-diastolic pressure of 10 mm Hg. Left ventricular developed pressure was significantly reduced in untreated hearts after myocardial infarction (120 ± 9 vs. 104 ± 5 mm Hg, $p < 0.05$, sham vs. myocardial infarction). This decrease of mechanical function was prevented by early (118 ± 9 vs. 113 ± 4 mm Hg, sham vs. myocardial infarction) but not by late bisoprolol therapy (119 ± 7 vs. 105 ± 6 mm Hg, $p < 0.05$, sham vs. myocardial infarction). Bisoprolol treatment was stopped 1 day before isolating the heart to avoid acute effects of beta-blocker treatment, and heart rate did not differ among the various groups.

Creatine kinase system. Creatine kinase activity of intact left ventricular tissue was reduced after infarction (6.4 ± 0.3 vs. 5.1 ± 0.2 IU/mg protein, $p < 0.05$; Fig. 1A). Bisoprolol treatment starting early preserved total CK activity at normal levels in infarcted hearts, whereas late treatment failed to prevent the decrease in CK activity. The relative distribution of the four CK isoenzymes BB, MB, MM and mitochondrial CK (mito-CK) changed after myocardial infarction (Fig. 1, B to E). The increase in the percentage of the BB isoenzyme after myocardial infarction in untreated hearts (2.2 ± 0.3% vs. 5.1 ± 0.4%, $p < 0.05$, sham vs. myocardial infarction) was reduced in the late treatment group to 3.3% and in the early treatment group of 2.4%. Similar changes were found for the MB isoenzyme: Bisoprolol reduced the increase of the MB isoenzyme completely (early treatment) or partially (late treatment). The MM percentage did not change in all groups. The significant decrease of mito-CK after infarction (34.8 ± 0.8% vs. 26.5 ± 1.2%, $p < 0.05$) was prevented by both late (33.0 ±

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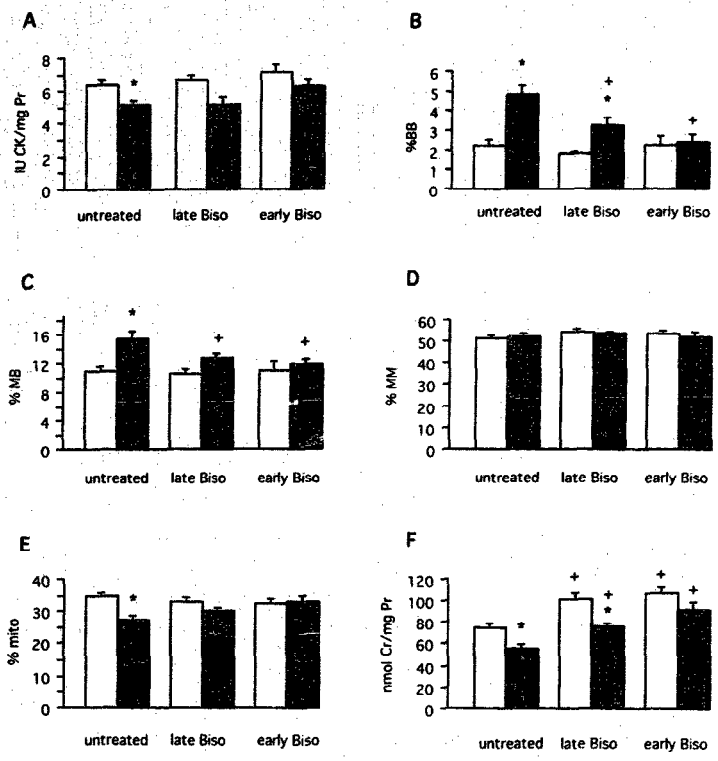

Figure 1. A to F, Effects of late and early bisoprolol (Biso) treatment on creatine kinase (CK) activity, CK-isoenzyme distribution and and total CK after myocardial infarction. *$p < 0.05$, myocardial infarction (**solid bars**) versus sham (**gray bars**); $^{+}p < 0.05$, treated versus untreated rats for n = 11 (sham untreated), 13 (myocardial infarction untreated), 12 (sham with late bisoprolol treatment), 13 (myocardial infarction with late bisoprolol treatment), 9 (sham with early bisoprolol treatment), 14 (myocardial infarction with early bisoprolol treatment). Cr = creatine; Pr = protein

1.2% vs. 30.3 ± 0.8%) and early treatment (32.5 ± 1.3% vs. 32.8 ± 1.9%). Thus, delayed bisoprolol therapy showed directionally similar but quantitatively smaller effects than early therapy on the CK system.

After myocardial infarction, total creatine content (Fig. 1F) decreased (74.2 ± 4.9 vs. 54.9 ± 3.3 nmol creatine/mg protein, $p < 0.05$, sham vs. myocardial infarction) in residual intact left ventricular myocardium. Early and late bisoprolol treatment completely prevented the reduction of creatine. Again, treatment starting immediately after infarction showed more substantial effects than late treatment. However, surprisingly, in bisoprolol-treated sham hearts, creatine content increased significantly above control levels of untreated sham hearts (74.2 ± 4.9) after early (107.7 ± 5.7 nmol creatine/mg protein, $p < 0.05$) and late treatment (101.1 ± 6.2 nmol creatine/mg protein, $p < 0.05$).

Lactate dehydrogenase and citrate synthase. Lactate dehydrogenase (LDH) activity was unaffected by myocardial infarction and bisoprolol therapy in untreated groups (0.94 ± 0.07 vs. 0.98 ± 0.08; sham vs. myocardial infarction), in late-treated groups (0.84 ± 0.04 vs. 0.75 ± 0.05) and early-treated groups (1.03 ± 0.07 vs. 0.97 ± 0.06). The anaerobic isoenzymes LDH4 (13.8 ± 0.9 vs. 20.0 ± 1.3, $p < 0.05$) and LDH5 (4.5 ± 0.3 vs. 7.8 ± 0.7, $p < 0.05$) increased significantly after myocardial infarction. This was completely prevented by early treatment (LDH4, 13.7 ± 1.1 vs. 13.7 ± 0.7; LDH5, 4.0 ± 0.3 vs. 5.2 ± 0.6); late treatment prevented the increase in part (LDH4, 12.7 ± 0.9 vs. 15.1 ± 0.8; LDH5, 4.8 ± 0.5 vs. 6.7 ± 0.6) (Fig. 2A–E). The activity of citrate synthase (IU/mg protein), a marker enzyme for mitochondrial mass, was comparable for all groups: 0.64 ± 0.04 versus 0.62 ± 0.03 (sham vs. myocardial infarction) in untreated groups, 0.71 ± 0.05 versus 0.73 ± 0.04 in late-treated groups and 0.80 ± 0.06 versus 0.74 ± 0.04 in early-treated groups (all p = NS, sham vs. myocardial infarction). Thus, mitochondrial mass estimated as citrate synthase activity did not change after myocardial infarction or by bisoprolol treatment.

Discussion

Hypertrophy. Quantitative histologic studies have shown that surviving myocytes after myocardial infarction increase in length and diameter (19). This is the morphologic substrate of the present biochemical studies. The fact that in our study left ventricular weight remained unchanged despite loss of myocardium from infarction and development of a thin scar suggests hypertrophy. However, left ventricular weight or the ratio of left ventricular weight to body weight is only a crude measure for hypertrophy and does not necessarily reflect the inhomogeneous changes at the cellular level. Previous studies

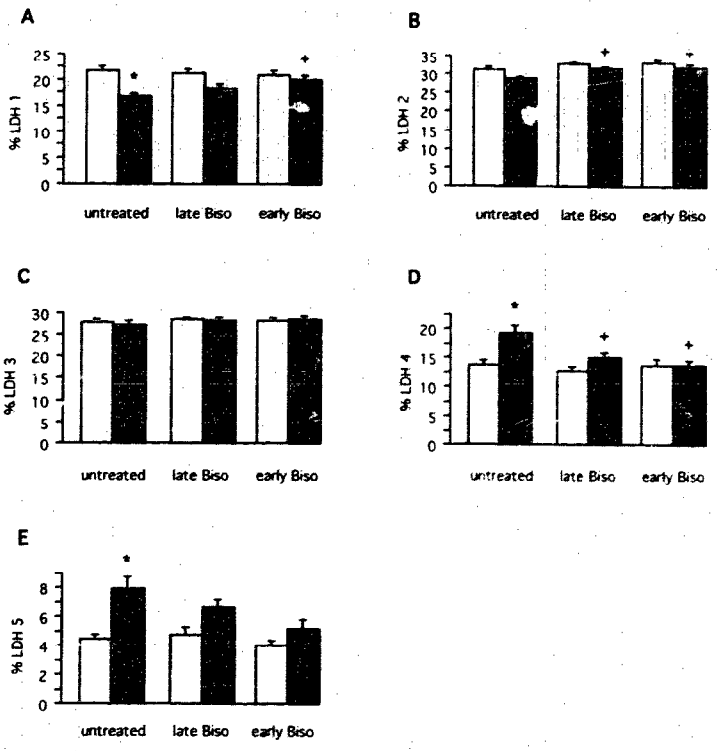

Figure 2. A to E, Effects of late and early bisoprolol (Biso) treatment on the distribution of the five lactate dehydrogenase (LDH) isoenzymes after myocardial infarction. *$p < 0.05$, myocardial infarction (**solid bars**) versus sham (**gray bars**); †$p < 0.05$ treated versus untreated rats for n = 11 (sham untreated), 13 (myocardial infarction untreated), 12 (sham with late bisoprolol treatment), 13 (myocardial infarction with late bisoprolol treatment), 9 (sham with early bisoprolol treatment), 14 (myocardial infarction with early bisoprolol treatment).

using the same rat infarct model have suggested that beta-blocker treatment blunts hypertrophy but promotes left ventricular dilation at the same time (20,21), with a net result of unchanged left ventricular weight after beta-blockade in infarcted hearts. Thus, bisoprolol may have had effects on morphology not reflected in the present study.

Mechanical function. Only limited data have been presented on functional effects of beta-blockade on surviving myocardium after myocardial infarction. Our data support the hypothesis that long-term beta-blocker treatment started early after myocardial infarction may in part prevent deterioration of mechanical function. Clearly, using developed pressure in a Langendorff preparation to measure left ventricular function has limitations. Loading conditions in vivo may be quite different, and geometric changes of the heart will greatly influence left ventricular function (13). Long-term beta-adrenergic blockade improved basal isometric contraction in noninfarcted isolated papillary muscle in rats after myocardial infarction, a model that eliminates the influence of geometric changes of left ventricle (3). Although these investigators founds an up-regulation of beta-adrenoreceptor density after propranolol treatment, it is unlikely to be the reason for improved mechanical performance because adenylate cyclase activity was still impaired (3). Mechanical function measured in vivo in rats post–myocardial infarction was not improved after long-term beta-blockade (21). In this latter study beta-blocker treatment was started 3 weeks after coronary ligation, similar to the late-treatment protocol in the present study, which did not prevent deterioration of left ventricular performance. Thus, the conflicting results may in part be explained by the different treatment regimens (early vs. late).

Creatine kinase and lactate dehydrogenase systems. The major result of the present study was that bisoprolol treatment prevented an increase in the anaerobic lactate dehydrogenase isoforms, the shift toward the fetal CK isoenzymes BB and MB and the depletion of creatine, which we previously described after myocardial infarction (10). During development from fetal to adult myocardium, total CK activity and mito-CK increase, whereas the fetal isoenzymes BB and MB decrease (22). The increase in fetal isoenzymes BB and MB is a uniform stress response observed after chronic coronary artery occlusion in the dog (23), in rat and dog facing pressure or volume overload or hypertension (24–26) and in myocardium of patients with valvular aortic stenosis or coronary artery disease (27). This isoenzyme shift was interpreted as an adaptive mechanism, because the affinity of the fetal B-type isoenzymes for creatine phosphate is higher than the affinity of the MM isoenzyme, making the phosphoryl transfer from creatine phosphate to adenosine triphosphate (ATP) more efficient. In the failing myocardium of 18-month old spontaneously hyper-

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tensive rat, reductions of total CK activity, mito-CK and total creatine occur in addition (24). Increases of anaerobic lactate dehydrogenase were also described in biopsy samples of patients with severe chronic heart failure (28).

Effects of beta-blockade on hypertrophy and on the adrenergic system might prevent these CK and lactate dehydrogenase changes. Chronic tissue hypoxia may be a possible trigger mechanism (27), or high serum catecholamine levels (29). In addition, beta-blocker treatment might have an energetically favorable effect by chronically reducing heart rate, rendering the adaptive increase of CK and lactate dehydrogenase isoforms unnecessary. Clearly, further studies are required to address the mechanism and the importance for myocardial function of this beta-blocker effect. The lactate dehydrogenase system changes are unlikely to be of significance for myocardial function, because lactate dehydrogenase is not a rate-limiting enzyme in the glycolytic pathway. This may, however, indicate prevention of a shift to chronic anaerobic metabolism.

Creatine content. Under bisoprolol therapy, infarcted hearts maintain normal creatine levels. However, even in sham-operated hearts, a substantial increase of creatine by long-term beta-blocker treatment was observed. Similar findings were reported by Chapados et al. (30) in healthy turkey hearts. Cardiomyocytes take up creatine from the bloodstream by specific plasma membrane creatine transport proteins (31). In isolated myoblasts up-regulation and down-regulation of this specific transporter maintained normal creatine levels despite varying extracellular creatine concentrations (32). Similar results were observed in rat heart tissue after creatine feeding (33). Therefore, an explanation for increased myocardial creatine levels by bisoprolol might be an effect on creatine transporter kinetics.

What might be the potential consequence of preservation of myocardial creatine in surviving myocardium after myocardial infarction? Because the ratio of phosphorylated to unphosphorylated creatine remains constant under all but acutely ischemic or hypoxic conditions, supplementation of intracellular creatine might preserve phosphorylated creatine and energy reserve in the surviving myocardium. However, the effect of beta-blockade on phosphocreatine has not been measured. In patients with severe dilated cardiomyopathy, creatine phosphate/ATP ratios measured by phosphorus-31 magnetic resonance spectroscopy increased to normal levels during clinical recompensation with treatment with digitalis, diuretic drugs, angiotensin-converting enzyme inhibitors and the beta-blocker metoprolol (34). It remains unclear whether energy metabolism changes were primary or secondary to hemodynamic improvement. Beta-blocker therapy may improve mechanical work without an increase in oxygen consumption in patients with severe heart failure (5,35). Thus, reduction of energetic costs of cardiac performance by beta-blockade may also contribute to preservation of energy reserve. At this point it remains speculative whether long-term beta-blocker treatment has direct effects on creatine transport and metabolism and myocardial enzymes or whether these changes are secondary, for instance, to a reduced heart rate. Either one would be important, however, and the data suggest so far unknown effects on beta-blockade in chronic cardiac dysfunction.

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